

Technical Data Sheet

MOLEQULE-ON®

Bismuth Sulfite Agar (Wilson Blair) USP

Cat #: MM-M-N011

Highly selective medium for the isolation of Salmonella spp, particularly Salmonella typhi, from clinical specimens.

Principles and uses:

Bismuth Sulfite Agar (Wilson Blair) is a modification of the Wilson Blair Medium, and generally accepted as routine for the detection of most Salmonella, in particular Salmonella typhi.

Peptone and beef extract provide nitrogen, vitamins, minerals and amino acids essential for growth. Dextrose is the fermentable carbohydrate providing carbon and energy, Bismuth sulfite indicator and brilliant green are inhibitors of Gram positive bacteria and members of the coliform group. Disodium phosphate acts as a buffer system and bacteriological agar is the solidifying agent.

Ferrous sulfate is included for detection of H₂S production. When H₂S is present, Salmonella spp reduces the iron salts to iron sulfate, which produces a black colony and turns the bismuth indicator to metallic bismuth, surrounding the area of the colonies with a bright sheen.

The colonies of S. typhi are black surrounded by a black or brownish zone, with a metallic sheen. In heavy growth areas, these may appear as light green colonies. Other strains of Salmonella produce black to green colonies with little or no darkening of the surrounding medium. Shigella spp, other than Shigella flexneri and Shigella sonnei, do not grow. Those colonies that do grow are brown to green, raised with a crater-like appearance. E.coli is partially inhibited, occasionally growing with brown or greenish glistening colonies. A few Enterobacter strains may grow with raised, mucoid colonies, having a silvery sheen lighter than S. typhi. Colonies of coliforms that produce H₂S form colonies similar in appearance to S. typhi. These may be readily differentiated as they produce gas with lactose media, e.g. TSI Agar (Cat.MM-M-046) or Kligler Iron Agar (Cat. MM-M-042). The hydrolysis of urea in Urea Broth (Cat. MM-M-226) or Urea Agar Base (Cat. MM-M-110) may be used to identify Proteus spp.

Formula per Litre:

Bacteriological agar	20g	Bacteriological peptone	10g
Brilliant green	0.025g	Dextrose	5g
Disodium phosphate	4g	Ferrous sulfate	0.3g
Beef extract	5g	Bismuth sulfite indicator	8g

Preparation:

Suspend 52.3 grams of medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. AVOID OVERHEATING. DO NOT AUTOCLAVE. Cool to 45°C (very important), mix well and dispense into plates.

Instructions for use:

For clinical diagnosis, the type of sample is stool.

- Inoculate Bismuth Sulfite Agar by streaking the surface to obtain isolated colonies.
- The pour plate inoculation method can be also used, mixing the sample with the liquid medium and allowing the plate to solidify.
- All plates are incubated 40- 48 hours at 35±2°C.
- The solidified plates should have a uniform, opaque, cream to pale green appearance.
- If kept in refrigeration, the medium will slowly oxidize. It is recommended to keep the plates refrigerated for 4 days before use to reduce inhibition and thus to be able to isolate Salmonella in less heavily contaminated samples.

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Quality control:

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
Flocculent precipitate	Fine powder	Light green	Opaque white with a green tint	7.5 ± 0.2

Microbiological test:

Inoculation conditions: (35±2 °C / 40-48 h).

Microorganisms	Specification	Characteristic reaction
Shigella flexneri ATCC 12022	Partial inhibition	Brown colonies
Salmonella enteritidis ATCC 13076	Good growth	Black colonies with bright metallic
Salmonella typhi ATCC 19430	Good growth	Black colonies with bright metallic
Escherichia coli ATCC 25922	Partial Inhibition	Brown-green colonies
Enterococcus faecalis ATCC 29212	Inhibition	

Storage:

Once opened keep powdered medium closed to avoid hydration.



Bibliography:

Wilson, W.J., and E.M. Blair 1.926 A combination of Bismuth and Sodium Sulfite affording an enrichment and selective medium for the typhoid-paratyphoid groups of bacteria. J. Pathol. Bactend 29:310.
United States Pharmacopoeia Convention 1.995. The United States Pharmacopoeia 23rd ed.